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RESEARCH ARTICLE

Comparative Study of Nutritional Composition and Rheological Behaviors of a set of Wild and Improved Cultivars of Tunisian Millet Flours

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ABSTRACT: Pearl millet is an important source of nutrients for human diet. In the present study, four wild and improved Tunisian pearl millet varieties were evaluated in terms of their nutritional composition and rheological behaviors. The nutritional composition screening revealed that millet had important nutrients quantities namely, starch (59.17- 75.54 %), protein (9.19-14.6 %), ash (2.48-2.56 %), lipid (5-6.31%), β-carotene (0,059-0,097 mg/100 g) and total phenolic (0.26-0.39%). Principal components analysis showed that IRA1 (improved) and Jerbi (wild) varieties had the richest nutritional value, while Magrebi (wild) had high mineral and amylopectin contents. However, IRA2 (improved) variety had high amount of proteins. The rheological evaluation showed that all millet starch and flour depicted shear-thinning (pseudoplastic) behavior ($n < 1$) fitting perfectly to the Herschel–Bulkley model (R^2 ranging from 0.98 to 0.99). These findings confirmed important genetic diversity between the studied seeds.

Key words: Pearl millet, starch, nutritional composition, shear-thinning behavior

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I.INTRODUCTION

One of the globally grown cereals is millet and it is characterized with different importance across continents and within regions of the world [1]. Millet includes pearl millet (Bajra; *Pennisetum glaucum*) and other minor millets such as Finger millet (Ragi; *Eleusine coracana*), Kodo millet (Arikalu; *Paspalum setaceum*), Proso millet (Cheena; *Penicum miliaceum*), Foxtail millet (Kauni; *Setaria italic*), Little millet (Kutki; *Panicum sumatrense*) and Barnyard millet (Sanwa; *Echinochloa utilis*) [2]. Pearl millet (*Pennisetum typhoideum*) is a staple food for population below poverty line for economic reasons [3]. Pearl millet has the properties of being tolerant to drought and heat; it is widely grown in tropical regions of the world including Africa and Asia [4]. Attempts have been made to combine high grain yield with good nutritional qualities [5].

Nutritional quality of food is a key element in maintaining human overall physical well-being because nutritional well-being is a sustainable force for health and development and maximization of human genetic potential [6]. The nutritional properties of pearl millet have received more attention than those of the other common millets [5]. Indeed, it constitutes an important source of dietary calories and protein in the daily diet of a large segment of the poor population [7]. It contains also abundant nutritional ingredients besides anti-nutrition factors, such as phytic acid, tannins, polyphenols and trypsin inhibitor substances that can reduce food digestibility [8]. Polyphenols have several properties, including antioxidants, anti-mutagenic, anti-oestrogenic, anti-carcinogenic and anti-inflammatory, antiviral effects and platelet aggregation inhibitory activity that might potentially be beneficial in preventing or minimizing the incidence of diseases [9].

Although the nutritive potential of millets in terms of protein, carbohydrate and energy values are comparable to the popular cereals like rice, wheat, barley or bajra [9], it is an underutilized cereal. As a matter of fact, this research focus was on the quantification of the nutrients that millet could provide to human diet. Indeed, nutritional assessment of millets properties will enhance the awareness of its consumption benefits. On the other hand, the evaluation of millet technological properties will certainly encourage its industrialization. Further, a comparative study of native and improved millet genotypes will enable a clear vision of the current quality of millet in Tunisia. Therefore, the objectives of the present study were to evaluate the approximate nutritional composition of wild and improved pearl millet commonly grown in Tunisia and to investigate their rheological behaviors for further industrial application.

II.MATERIAL AND METHODS

Grains samples and preparation

Grain samples of millet seeds (*Pennisetum glaucum* L.) were kindly obtained from IRA Médenine (Institute of Arid Regions Médenine, Tunisia). IRA's collection team established a collection of pearl millet (*Pennisetum glaucum*) germplasm from the arid regions of Tunisia (landraces and modern lines) as vigorously described by [10].

In order to study the impact of breeding programs on nutritional and rheological properties of pearl millet, four Tunisians cultivars were used; two wild populations named, Jerbi and Magrebi and two improved lines named, IRA1 and IRA2. The seeds were cleaned, milled into fine flour and sieved through a 0.250 mm mesh. The samples were stored at 4 °C for further analysis and used within 2 weeks of milling.

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Nutritional composition

The millet flour samples were assessed for their nutritional composition mainly, carbohydrates, proteins, lipids, ash, β-carotene, and polyphenols.

Total starch and amylose contents were determined as previously described by Walker and Jarvis (1993) [11]. The quantification was performed with the use of the colorimetric determination of starch with iodine. Protein was quantified following the procedure of Kruger (1994) [12]. Protein concentration was measured using a standard calibration curve with bovine serum albumin. *Lipid content* was determined after extraction at 60 °C for 8 h using petroleum ether by Soxhlet method. β-carotene were extracted using the method described by Liu et al. (2004) [13]. β-carotene was used as standard, and β-carotene content of samples was expressed as mg of β-carotene equiv/100 g of sample. Ash content was determined according to the standard method ISO 2171. The assay of total phenols was carried out using the method of Singleton and Rossi (1965) [14]. Gallic acid was used as standard, and the total phenolic content of samples was expressed as mg of gallic acid equiv/100 g of sample.

Starch isolation

Isolation of starch was carried out as described by Lawal (2002) [15] with some modifications. In brief, 100 g grains were steeped into 200 ml of 0.25 % NaOH solution for 24 h at 4 °C. Then, grains were vigorously washed under a stream of water. The cleaned grains were ground in a blender for 10 min with distilled water (1:1 v/v). The recovered suspension was resuspended in 700 ml of distilled water and sieved through muslin. After centrifugation (2000 rpm, 30 min), the recovered starch was suspended in 200 ml of distilled water and 4 ml of toluene. The mixture was stirred for 30 minutes. The top layer emulsion was scraped and the mixture was allowed to settle for two hours. After removing the supernatant, the samples were dried in a convective oven at 40 °C for 24 h. The extraction yield is expressed as a percentage of final starch weight relative to the initial grains weight.

Rheological properties of millet flour and starch

Gel preparation

Millet flours and starches dispersions were used for the preparation of gels at different concentrations (10 % and 5 %, respectively). Gels were obtained after heating the dispersions at 100 °C for 4 min with continuous stirring in a magnetic stirrer.

Rheological measurements

Rheological behavior of millet flour and starch gels was assessed using a coaxial rheometer (AR-G2, TA instruments, USA). A temperature of 30 °C was maintained constant during the rheological measurement. Shear-stress and shear-rate data were fitted to the Herschel-Bulkley rheological model ($\sigma = \sigma_0 + k \times \varepsilon^n$); where, σ is shear-stress (Pa); ε is shear-rate (s^{-1}), k is fluid consistency index ($Pa.s^n$), n is flow behavior index, and σ_0 is the yield stress (Pa).

Statistical analysis

Experimental data were assessed by analysis of variance (ANOVA). Significant differences among the mean values were calculated using Duncan's test with probability $P<0.05$. Principal component analysis (PCA) was performed based on correlation matrix. Statistical analysis was accomplished using the SPSS version 13.0 (SPSS Inc., Chicago, IL, USA).

III.RESULTS AND DISCUSSION

Nutritional composition of millets flours

Proteins, carbohydrates, minerals and vitamins are some of the basic constituents of the consumable part of plant which determines its nutritional [1]. Table 1 presented an approximate nutritional composition of wild and improved millet cultivars. Statistical results revealed that the genotype had a high significant effect on all the studied components, exception of β-carotene.

Starch is cereals main carbohydrates. In fact, cereal crops accumulate starch in the seed endosperm as an energy reserve [16]. Table 1 shows that total starch content of the four studied varieties, varied from 59.17% to 75.54 %. These findings are within the range of Sudanese pearl millet (58-70%) as previously reported by Abdalla et al, (1998) [17]. The improved line, IRA1, had the highest total starch content (75.54%). The average value of starch content of improved varieties was higher than landraces. This result could be attributed to the tendency of new breeding programs to increase carbohydrates content. However, Jerbi (wild), recorded high starch content as compared to IRA2. In this case, Jerbi (wild) should be preserved and considered as a good candidate for selection knowing that landraces are well-adapted to arid conditions, poor soils, brackish [10]. Regarding pearl millet amylose content, its variability range was from 15.93 to 19.88% (mean value = 17.9 %). As expected, modern lines scored the highest amylose content (19.79 %) as compared to landraces (17.47%). In particular, IRA2 and IRA1 amylose contents (19.88% and 19.70 %, respectively) were slightly higher than the wild line Jerbi (19.01 %).

Millets proteins are good sources of essential amino acids except lysine and threonine but are relatively high in methionine [6]. Protein content ranged from 9.19 to 14.6 %. IRA2 recorded the highest protein content (14.6 %), while IRA1 recorded the lowest value (9.19%) indicating important diversity. As for wild genotypes, Magrebi and Jerbi had medium values (12.87% and 10.42%, respectively). These results were in consistence with the works of Suma and Urooj (2014) [3] on Indian pearl millet varieties namely, Kalukombu and Maharashtra Rabi Bajra (9.3 and 10.2, respectively). It could be concluded that breeding programs impact on protein content was less pronounced than starch content. The ash content oscillated from 2.48 % to 2.56 %. The local population, Magrebi, had the highest value (2.56%). Lipids are relatively minor constituents in cereals grains; whereas they contribute significantly in diet as a source of invisible fat and essential fatty acids [18]. Lipid content of millet samples varied from 5 to 6.31%. IRA1 had the highest content of lipid (6.31%). In contrast, IRA2 had the lowest content (5.05%). It was noted also that Jerbi recorded high content of lipid (6.13 %). This result was within the range 2.7–7.1% previously reported by Abdalla et al. (1998) [17]. As a result, high content of fat in millet should be taken into consideration during storage and processing [19]. Carotenoids are important biological precursors of vitamin A and are generally believed to enhance human health [20]. The average content of β-carotene in the present study was 0.059 mg/100g. Thus, the consumption of millet could be considered a possible cereal source of vitamin A. As a matter of fact, IRA2 had the highest content (0.097 mg/100 g). Likewise, wild genotype, Jerbi had important content of β-carotene (0.088 mg/100g). However, Magrebi (wild line) recorded the lowest content (0.059 mg/100g). Millets contain phytochemicals that may provide beneficial health effects other than basic nutrients [21]. Total phenolic content varied from 0.26 to 0.39%. Jerbi had the highest total phenol content (0.39%), contrary to Magrebi (0.26%). Besides, IRA1 and IRA2 presented a phenolic medium content (0.35% and 0.30%, respectively). These findings were in concordance with the work of Nambiar, et al., (2012) [22] on pearl millet phenols (0.27 – 0.42 %). Thus, it may be concluded that pearl millet could be considered a possible source of phytochemicals. Principal components analysis (PCA) was performed for a better understanding of varietal selection influence on approximate composition of Tunisian millet. Figure 1 illustrated the biplots of the first two axes of the PCA. The first principal component (PC1) accounted for 51.76 % of the total variability, which was explained by amylose, amylopectin, starch, ash and β-carotene contents; whereas the second component (PC2) accounted for 39.61 % mainly explained by lipid, protein and phenols contents. The PCA biplot (Figure 1b) established three distinct groups. The first group comprised IRA1 and Jerbi. Moreover, IRA2 and Magrebi varieties made two distinct groups. We projected also the average values of wild and improved millets cultivars. They made intermediate groups. Thus, PCA results made clear the nutritional components distribution between the studied lines. As a result, IRA1 and Jerbi varieties had the richest nutritional value suggesting a genetic resemblance between both lines. However, Magrebi variety was characterized by high mineral content and amylopectin. Although IRA2 variety had high amount of proteins, it had also a midway location between both groups indicating its intermediate nutritional composition.

Rheological behaviors of millet fours and starches

The rheological behaviors of millet gels (flour gel and starch gel) are presented in figures 2 and 3. Results showed that the shear stress of the samples increased with the increase in shear rate. Moreover, the plot of shear stress (Pa) vs shear rate (s^{-1}) obtained for millet flour gels and starch gels samples were not linear and therefore they are non-Newtonian fluids. This behavior could be explained by the molecules structural breakdown generated by the rotating measurement system and the increased constituent alignment [23]. Figures 2 and 3 revealed also that starch gels presented lower values of shear stress as compared to flour gels values.

Table 1: Nutrient composition of pearl millet

Geno type	Total Starch (%)	Amylose (%)	Amylopectin (%)	Protein (%)	Ash (%)	Lipid (%)	B-carotene (mg/100g)	Polyphenols(%)
Jerbi	62.41c	19.01 b	80.99 b	10. 42 b	2.45a	6.13 c	0.088	0.39 d
Magrebi	59.17 a	15.93 a	84.07 c	12.78 c	2.65 d	5.55 b	0.059	0.26 a
Wild	60.79	17.47	82.53	11.60	2.55	5.84	0.074	0.33
IRA1	75.54 d	19.70 c	80.30 a	9.19 a	2.48 b	6.31 d	0.097	0.35 c
IRA2	60.72 b	19.88 d	80.12 d	14.60 d	2.51 c	5.05 a	0.080	0.30 b
Improved	68.13	19.79	80.21	11.90	2.50	5.68	0.089	0.33

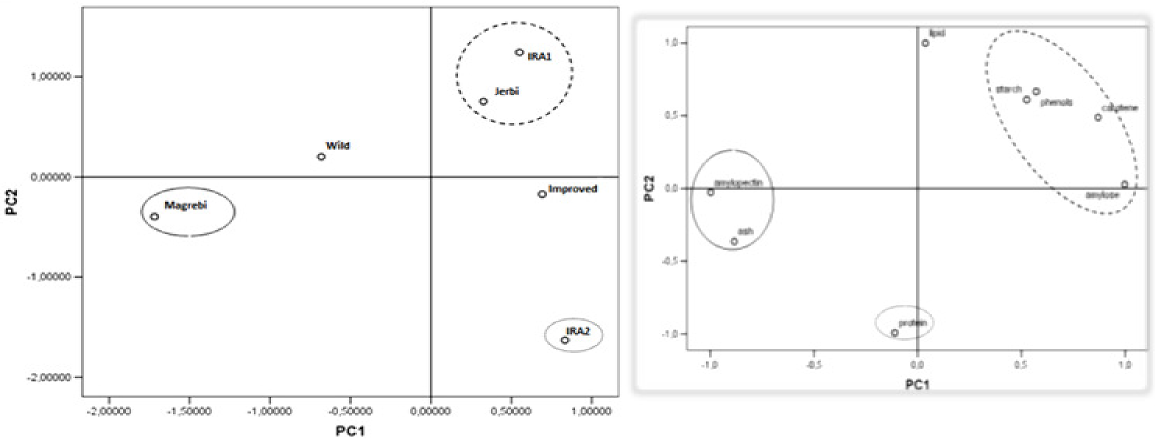

Figure 1: Principal components biplots of the approximate composition of Tunisian pearl millet grains ;(a): Biplot of principal component analysis, (b): Rotated principal scores of the millet genotypes projected onto the plane of the first and second principal components.

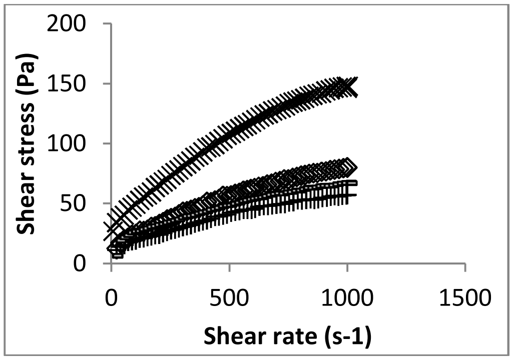

Figure 2: Flow curves of millet flour at 30 °C. (-: Jerbi; ×: Magrebi ; +: IRA1; ◊: IRA2).

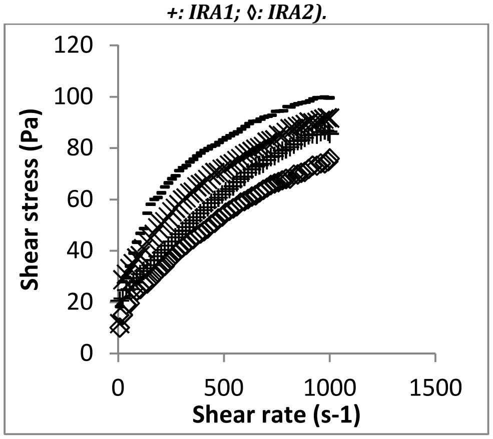

Figure 3: Flow curves of millet starches at 30 °C (-: Jerbi; ×: Magrebi ; +: IRA1; ◊: IRA2).

The Herschel–Bulkley model can be used to describe the rheological behavior of certain non-Newtonian fluids [24]. This model fitted perfectly to the above presented experimental data (R^2 ranging from 0.98 to 0.99). Table 2 presented the parameters of Herschel-Bulkley model relative to flour and starch gel samples. All starch and flour gel samples depicted shear-thinning (pseudoplastic) behavior ($n < 1$). The yield stress showed important variability with a range of 6.6 to 27 Pa (see table 2). Magrebi variety had the highest fluid consistency index (3.48), while the improved line IRA1 had the lowest value (0.11). Flow behavior index value of IRA1 variety was slightly lower than 1, whereas Magrebi variety had the lowest value (0.456). In fact, it could be deduced that IRA1 gel shear-thinned more than Magrebi gel. On the other hand, fluid consistency index of the wild line Jerbi starch was the highest indicating its high consistency which could be probably attributed to starch content and structure. Rheological assessment revealed important diversity between wild and improved genotypes. Moreover, Loumerem et al. works (2008) [10] gave special interests to the pearl millet landraces of Tunisian dry regions confirming the high phenotypical variability. These findings could explain the important genetic diversity between the two studied wild lines rheological behaviors.

IV. CONCLUSION

The current research purpose was to nutritionally and technologically evaluate improved and wild millets accessions cultivated in Tunisia.

Table 2: Herschel-Bulkley model parameters obtained for millet flours and starches gels

	Herschel-Bulkley model parameters							
	σ_0 (Pa)		**K** (Pa.s^n)		**n**		**R^2**	
	Starch	**Flour**	**Starch**	**Flour**	**Starch**	**Flour**	**Starch**	**Flour**
Jerbi	6.6 a	18.3 c	2.63 d	2.30 c	0.451 a	0.533 b	0.980	0.997
Magrebi	27 d	11.6 b	0.70 c	3.48 d	0.756 b	0.456 a	0.986	0.993
IRA1	11.3 b	20.6 d	0.18 a	0.11 a	0.811 d	0.949 d	0.991	0.995
IRA2	12.7 c	10.1 a	0.32 b	0.74 b	0.787 c	0.654 c	0.992	0.989

Data are expressed as mean from three replicates. Means of same column followed by different letters differ significantly ($p < 0.001$).

Statistical data revealed significant differences between wild and improved millet. Further, PCA confirmed the biodiversity among the studied millet samples in terms of nutrient contents. PCA deemed also that Jerbi (wild) was characterized by a good nutritional composition in comparison with IRA2 (improved). Rheological assessment was performed in order to look into millet flour and starch technological properties. Although results showed that all the obtained gels had a pseudoplastic behavior, relevant differences was recorded between the samples as a function of breeding history.

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